REMARKS

Claims 1-9, 11-18, 20-28, and 39-44 are pending. Claim 18 has been amended without prejudice or disclaimer. No new matter has been added.

Withdrawn Rejections

Applicants thank the Examiner for withdrawing the 35 U.S.C. § 103 rejections that were pending in the previous Office Action.

35 U.S.C. § 103

Ladner, Anderson, and Chandrashekar. The Office at pages 3-8 of the Action maintains its position that claims 1-9, 13, 15-17, 20-27, and 39-44 are allegedly obvious in light of Ladner (U.S. Pat. No. 5,403,484; hereinafter "the '484 patent"), Anderson (U.S. Pat. No. 6,649,419; hereinafter "the '419 patent"), and Chandrashekar (U.S. Patent No. 5,854,051; hereinafter "the '051 patent").

Ladner, Anderson, Chandrashekar, and Janda. At pages 8-10, the Office alleges that claims 1-10, 12-17, 20-28, and 39-44 are obvious in light of the '484 patent, the '419 patent, the '051 patent, and Janda (U.S. Pat. No. 5,571,681; hereinafter "the '681 patent").

The Office does not set forth any grounds of rejection that rely on the '051 patent. Instead, the Office only sets forth a rejection based on Ladner, Anderson, and Janda. As the rejection based on these three references was withdrawn in the present Office Action, Applicants assume that the Office meant to rely the '051 patent in support of the present rejection. In an effort to expedite prosecution, Applicants will address this rejection and address the '051 patent, as it is relied on in the above-noted rejection.

However, if the present Reply does not overcome this rejection, Applicants submit that any subsequent Office Action cannot be made final, as the Applicants have not been afforded an opportunity to address the merits of a rejection based on Ladner, Anderson, Janda, and Chandrashekar (the '051 patent).

Ladner, Anderson, Chandrashekar, and McCafferty. The Office at pages 10-11 alleges that claims 1-9, 13, 15-18, 20-27, and 39-44 are obvious in light of the '484 patent, the '419 patent, the '051 patent, and McCafferty (U.S. Pat. No. 5,969,108; hereinafter "the '108 patent").

Ladner, Anderson, Chandrashekar, Janda, and Steinbuchel. At pages 11-12 of the Action, the Office alleges that claims 1-17, 20-28, and 39-44 are obvious in light of the '484 patent, the '419 patent, the '051 patent, the '681 patent, and Steinbuchel (U.S. Pat. No. 6,022,729, hereinafter "the '729 patent").

The Office does not set forth any grounds of rejection that rely on the '051 patent. Instead, the Office only sets forth a rejection based on Ladner, Anderson, Janda, and Steinbuchel. As the rejection based on these four references was withdrawn in the present Office Action, Applicants assume that the Office meant to rely the '051 patent in support of the present rejection. In an effort to expedite prosecution, Applicants will address this rejection and address the '051 patent, as it is relied on in the above-noted rejection.

However, if the present Reply does not overcome this rejection, Applicants submit that any subsequent Office Action cannot be made final, as the Applicants have not been afforded an opportunity to address the merits of a rejection based on Ladner, Anderson, Janda, Steinbuchel, and Chandrashekar (the '051 patent).

Applicants' Reply.

Applicants respectfully disagree with the Office's positions. The claims recite, in part:

> - A method of selecting phage that encode a target binding protein from a plurality of display phage. The method includes the step of producing replicate phage from the infected cells in the presence of the target immobilized to the support, thereby forming replicate phage immobilized to the support via binding to the target, wherein the producing is completed in less than 4 hours (e.g., claim 1);
>
> - A method of identifying members of a bacteriophage library that have a desired binding property. The method includes the step of amplifying members of a subset of bacteriophage members in less than 4 hours (e.g., claim 20); and
>
> - A method of selecting a nucleic acid that encodes a binding protein from a library of display phage. The method includes the step of producing phage from the infected cells in the presence of the target, the produced phage being replicates of

> phage that bind to the target, wherein the producing is completed in less than 4 hours (e.g., claim 24).

The methods recite that the producing steps are completed in less than 4 hours, or that less than 4 hours elapse in the amplifying steps.

The cited references, considered alone or in combination, fail to render such methods obvious.

In Applicants' Reply filed on September 22, 2009, the obviousness rejections were addressed with respect to the '484 patent, the '419 patent, the '681 patent, the '108 patent, and the '729 patent. Applicants reiterate the remarks made in the Reply filed on September 22, 2009 (and the Declaration submitted therewith) with respect to those references.

As indicated in the present Office Action, Applicants overcame the obviousness rejections based on these references as the present Office Action withdrew each of those rejections.

In the present Office Action, the Office newly cites the '051 patent to support its position that completing producing steps in less than 4 hours are obvious, and/or that less than 4 hours elapsing in amplifying steps is obvious. The Office alleges in part:

> One experimental approach for expressing the desired proteins from the library of nucleic acids involves a 4 hour incubation of the phage (see Example 4, paragraph bridging cols. 28 and 29, <u>especially col. 29 lines 1-4</u> [of the '051 patent]). (Office Action at page 8; emphasis in the original)

The Office further alleges at page 8:

> Regarding the time limitations such as "4 hours" or less time as in claims 39-44, such adjustments are merely routine experimental parameters as demonstrated by Chandrashekar with predictable outcomes (i.e., lower numbers of colonies).

Applicants submit that the Office has mischaracterized the disclosure of the '051 patent.

As an initial matter, the cited passage at Example 4 of the '051 patent describes a <u>seven hour</u> incubation of the phage:

> Briefly, phage were plated onto a lawn of *E. coli* XL1-Blue MRF' (available from Stratagene) at a density of $25x10^3$ phage per petri dish (150 mm^2) and grown at 37º C. **for 4 hr**. When plaques were visible, isopropyl-β-D thiogalactoside (IPTG)-impregnated

> nitrocellulose filters were placed on the plates **for 3 hr** at 37º C. (col. 28, line 66 to col. 29, line 5; emphasis added)

As this passage makes clear, the phage were plated on a lawn of bacteria, incubated for four hours, filters were then placed on the plates and the plates were incubated for an additional three hours. This results in a total incubation time of seven hours.

Further, the three hours of incubation after the addition of IPTG is required to induce protein expression from infected bacteria and thus cannot be omitted. As described in Example 4 of the '051 patent, the methods describe an IPTG-inducible system.

Next, the experiments described at Example 4 of the '051 patent take over 12 hours to complete. For example, as stated in Example 4 of the '051 patent:

> A DiASNase nucleic acid molecule of 1753 nucleotides, herein referred to as $nDiASNase_{1753}$, was cloned from a *D. immitis* larval cDNA library by immunoscreening. Specifically, a *D. immitis* 48-hr L3 cDNA expression library was constructed in Uni-ZAP^{TM} XR vector (available from Stratagene Cloning Systems, La Jolla, Calif.), using a ZAP-cDNA Synthesis Kit (available from Stratagene) and 48-hr L4 mRNAs. The library was immunoscreened using the IgG-enriched anti-cuticle antisera described in Example 1, and **standard immunoscreening procedures** as described, for example, in Sambrook et al., ibid. Briefly, phage were plated onto a lawn of *E. coli* XL1-Blue MRF' (available from Stratagene) at a density of $25x10^3$ phage per petri dish (150 mm^2) and grown at 37º C. for 4 hr. When plaques were visible, isopropyl-β-D thiogalactoside (IPTG)-impregnated nitrocellulose filters were placed on the plates for 3 hr at 37º C. The filters were then removed and washed in 0.01% M phosphate-buffered saline, pH 7.4 with 0.05% Tween 20 (PBS/T), and then blocked in PBS/T containing 5% nonfat dry milk for one hr at room temperature. The filters were then incubated for 3 hr in mouse IgG-enriched anti-cuticle antisera, diluted 1:200 in PBS/T, that had been previously absorbed with *E. coli* antigens. Antibody reactivity with recombinant proteins was revealed by incubation of the filters with alkaline phosphatase-conjugated goat anti-mouse IgG antibodies (available from Kirkegaard and Perry) for 1 hr, followed by development with 5-bromo-4-chloro-3-indolyl phosphate/nitroblue tetrazolium (BCIP/NBT, available from Sigma). Clones that were reactive with the sera were selected and purified by **repeated cycles** of immune selection. (col. 28, line 56 to col. 29, line 19; emphasis added)

As this passage makes clear, standard immunoscreening procedures were used to isolate a nucleic acid encoding a particular enzyme (DiASNase). One cycle of the process described in this example takes over 12 hours to complete:

Step	**Duration**
Grow bacteria after phage added	4 hours
Continue growing bacteria after IPTG is added	3 hours
Remove filter and wash	>0 (no time is stated but must occur for some duration)
Block filter in PBS/T that contains dry milk	1 hour
Incubate filter in mouse IgG-enriched antisera	3 hours
Incubate with goat anti-mouse IgG antibodies	1 hour
Develop with BCIP/NBT	>0 (no time is stated but must occur for some duration)
Total	Over 12 hours

In contrast, the methods recited in the claims can be completed in shorter time than the methods described in the cited references. As described in the Declaration filed with the Reply of September 22, 2009:

6. In my opinion, the advantages provided by the methods recited in the amended claims are significant. For example, as described in the application, using methods known in the art, a single round *of selection takes* one to five days *to complete (see, e.g., par. bridging pages 1-2). As a result, if multiple rounds of selection are performed, the process would take well over a week. In large part, this is because such methods require that after selection against a target, phage that bind to the target are used to infect host cells and the phage-infected cells are grown for long periods of time, typically overnight. After the overnight growth, the phage must be pooled and/or purified before being used for another round of selection. Pooling and/or purifying steps further lengthen the total time needed to complete the selection procedure.*

In contrast, the claimed methods allow for multiple rounds of selection to be completed in a single day (see, e.g., page 15, first full par.) …

Applicants submit that, for at least these reasons, claims 1, 20, and 24 (and their rejected dependencies) are non-obvious over the combinations of cited references.

Withdrawal of these rejections is respectfully requested.

CONCLUSION

Applicants respectfully submit that all of the pending claims are in condition for allowance, which action is expeditiously requested. Applicants do not concede any positions of the Examiner that are not expressly addressed above, nor do Applicants concede that there are not other good reasons for patentability of the presented claims or other claims.

If this response is not considered timely filed and if a request for an extension of time is otherwise absent, Applicants hereby request any necessary extension of time. If there is a fee occasioned by this response, including an extension fee, please charge any deficiency to Deposit Account No. 50/2762.

Respectfully submitted,
Ladner et al., *Applicants*

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